IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re the application of: Hermann Bujard, and Manfred Gossen

Serial No.: N/A

Filed: June 4, 2001
(Continuation of Serial No. 09/161,902)

For: *TRANSGENIC ORGANISMS HAVING TETRACYCLINE-REGULATED TRANSCRIPTIONAL REGULATORY SYSTEMS (as amended)*

Attorney Docket No.: BBI-009C3CN2

Group Art Unit:

Examiner:

Commissioner for Patents
Box Patent Application
Washington, D.C. 20231

CERTIFICATION UNDER 37 CFR 1.10

Date of Deposit: June 4, 2001 Mailing Label Number: **EL 589 739 235 US**

I hereby certify that this 37 CFR 1.53(b) request and the documents referred to as attached therein are being deposited with the United States Postal Service on the date indicated above in an envelope as "Express Mail Post Office to Addressee" service under 37 CFR 1.10 and addressed to the Commissioner for Patents, Box Patent Application, Washington, D.C. 20231.

Larry Taylor
Name of Person Mailing Paper

Larry Taylor
Signature of Person Mailing Paper

PRELIMINARY AMENDMENT

Dear Sir:

Prior to examination of the above-identified application, please amend the application as follows:

In the Specification:

At page 1, line 3, and page 96, line 2, please delete the original title and insert the following new title: --Transgenic Organisms Having Tetracycline-Regulated Transcriptional Regulatory Systems--.

At page 1, line 6, under the heading "Related Applications", please delete lines 6-14, and insert the following:

--This application is a continuation of U.S. Serial No. 09/161,902, filed September 28, 1998, pending, which is a continuation of U.S. Serial No. 08/487,472, filed June 7, 1995, now U.S. Patent No. 5,912,411, which is a continuation-in-part of U.S. Serial No. 08/383,754, filed February 3, 1995, now U.S. Patent 5,789,156, which is a continuation-in-part of U.S. Serial No. 08/275,876, filed July 15, 1994, now U.S. Patent 5,654,168, which is a continuation-in-part of U.S. Serial No. 08/270,637, filed July 1, 1994, now abandoned. U.S. Serial No. 08/487,472 is also a continuation-in-part of U.S. Serial No. 08/260,452, filed June 14, 1994, now U.S. Patent 5,650,298, which is a continuation-in-part of U.S. Serial No. 08/076,327, filed June 14, 1993, now abandoned. U.S. Serial No. 08/487,472 is also a continuation-in-part of Serial No. 08/076,726, filed June 14, 1993, now U.S. Patent 5,464,758. The entire contents of each of these applications are incorporated herein by reference.--

At page 1, line 34, after "species such as", please delete "*E.coli*" and insert --*E. coli*--.

At page 2, line 20, before "or pleiotropic effects", please delete "cytoxicity" and insert --cytotoxicity--.

At page 4, line 32, please delete "Figure 9A-B" and insert --Figure 9A-9B--.

At page 4, line 33, please delete "Panel A" and insert --Figure 9A--.

At page 4, line 35, please delete "Panel B" and insert --Figure 9B--.

At page 8, line 19, after "sufficient to", please delete "acheive" and insert --achieve--.

At page 8, line 26, after "outside the", please delete "tetracyline" and insert --tetracycline--.

At page 8, line 29, after "the conserved", please delete "tetracyline" and insert --tetracycline--.

At page 8, line 34, after "of which", please delete "theTn10-" and insert --the Tn10--.

At page 11, line 8, after "indirect", please delete "mechanims" and insert --mechanism--.

At page 12, line 29, after "protein with an", please delete "endogeneous" and insert --endogenous--.

At page 13, line 5, after "non-limiting", please delete "exampleof" and insert --example of--.

At page 20, line 24, after "flexibility in the", please delete "permissable" and insert--permissible--.

At page 21, line 4, after "flexibility in the", please delete "permissable" and insert --permissible--.

At page 21, line 22, after "homologous recombinant", please delete "organims" and insert --organism--.

At page 21, line 25, after "expression of", please delete the second "the".

At page 23, line 32, after "fertilized", please delete "ooctye" and insert --oocyte--.

At page 26, line 21, before "with this target", please delete "conjuction" and insert --conjunction--.

At page 27, line 3, after "on two", please delete "seperate" and insert --separate--.

At page 29, line 25, after "second nucleic", please delete "acide" and insert --acid--.

At page 55, line 10, after "or absence of", please delete "tetracyline" and insert

--tetracycline--.

At page 56, line 6, after "Tc or", please delete "doxycyline" and insert --doxycycline--.

At page 57, line 1, after "luciferase", please delete "activitv" and insert --activity--.

At page 58, line 25, after "activity by", please delete "cultureing" and insert --culturing--.

In the claims

Please cancel claim 1.

Please add new claims 21-36, as follows:

--21. A transgenic organism having a transgene integrated into the genome of the organism and also having a *tet* operator-linked gene in the genome of the organism, wherein:

the transgene comprises a transcriptional regulatory element functional in cells of the organism operatively linked to a polynucleotide sequence encoding a fusion protein which activates transcription of said *tet* operator linked gene,

the fusion protein comprises a first polypeptide which is a mutated Tet repressor that binds to a *tet* operator sequence in the presence of tetracycline or a tetracycline analogue operatively linked to a second polypeptide which activates transcription in eukaryotic cells,

said *tet* operator-linked gene confers a detectable and functional phenotype on the organism when expressed in cells of the organism,

said transgene is expressed in cells of the organism at a level sufficient to produce amounts of said fusion protein that are sufficient to activate transcription of the *tet* operator-linked gene; and

in the presence of tetracycline or a tetracycline analogue in the organism, said fusion protein binds to the *tet* operator-linked gene and activates transcription of the *tet* operator linked gene such that the *tet* operator-linked gene is expressed at a level sufficient to confer the detectable and functional phenotype on the organism, wherein the

level of expression of the *tet* operator-linked gene can be downmodulated by depleting tetracycline or a tetracycline analogue from the organism.

22. A transgenic organism having a transgene integrated into the genome of the organism and also having a *tet* operator-linked gene in the genome of the organism, wherein:

the transgene comprises a transcriptional regulatory element functional in cells of the organism operatively linked to a polynucleotide sequence encoding a fusion protein which inhibits transcription of said *tet* operator linked gene,

the fusion protein comprises a first polypeptide which is a mutated Tet repressor that binds to a *tet* operator sequence in the presence of tetracycline or a tetracycline analogue operatively linked to a second polypeptide which inhibits transcription in eukaryotic cells,

said *tet* operator-linked gene confers a detectable and functional phenotype on the organism when expressed in cells of the organism,

said transgene is expressed in cells of the organism at a level sufficient to produce amounts of said fusion protein that are sufficient to inhibit transcription of the *tet* operator-linked gene; and

in the presence of tetracycline or a tetracycline analogue in the organism, said fusion protein binds to the *tet* operator-linked gene and inhibits transcription of the *tet* operator linked gene, wherein the level of expression of the *tet* operator-linked gene can be upregulated by depleting tetracycline or a tetracycline analogue from the organism.

23. A transgenic organism having a transgene integrated into the genome of the organism and also having a *tet* operator-linked gene in the genome of the organism, wherein:

the transgene comprises a transcriptional regulatory element functional in cells of the organism operatively linked to a polynucleotide sequence encoding a fusion protein which inhibits transcription of said *tet* operator linked gene,

said fusion protein comprises a first polypeptide that is a Tet repressor, operably linked to a heterologous second polypeptide which inhibits transcription of said *tet* operator-linked gene in eucaryotic cells,

said *tet* operator-linked gene confers a detectable and functional phenotype on the organism when expressed in cells of the organism,

said transgene is expressed in cells of the organism at a level sufficient to produce amounts of said fusion protein that are sufficient to inhibit transcription of the *tet* operator-linked gene; and

in the absence of tetracycline or a tetracycline analogue in the organism, said fusion protein binds to the *tet* operator-linked gene and inhibits transcription of the *tet* operator linked gene, wherein the level of expression of the *tet* operator-linked gene can be upregulated by administering tetracycline or a tetracycline analogue to the organism.

24. The organism of claim 21, wherein the mutated Tet repressor has at least one amino acid substitution compared to a wild-type Tet repressor.

25. The organism of claim 24, wherein the mutated Tet repressor is a mutated Tn10-derived Tet repressor having an amino acid substitution at at least one amino acid position selected from the group consisting of position 71, position 95, position 101 and position 102.

26. The organism of claim 24, wherein the mutated Tn10-derived Tet repressor comprises an amino acid sequence shown in positions 1 to 207 of SEQ ID NO: 2.

27. The organism of claim 21, wherein the second polypeptide of the fusion protein comprises a transcription activation domain of herpes simplex virion protein 16.

28. The organism of claim 22, wherein the second polypeptide of the fusion protein comprises a transcriptional silencer domain of a protein selected from the group consisting of v-erbA, the Drosophila Krueppel protein, the retinoic acid receptor alpha, the thyroid hormone receptor alpha, the yeast Ssn6/Tup1 protein complex, the Drosophila protein even-skipped, SIR1, NeP1, the Drosophila dorsal protein, TSF3, SF1, the Drosophila hunchback protein, the Drosophila knirps protein, WT1, Oct-2.1, the Drosophila engrailed protein, E4BP4 and ZF5.

29. The organism of claim 21, wherein expression of the transgene is regulated by at least one *tet* operator sequence.

30. The organism of claim 21, wherein expression of the transgene is regulated by at least one virally-derived regulatory element.

31. The organism of claim 21, wherein expression of the transgene is regulated by at least one tissue-specific regulatory element.

32. The organism of claim 21, wherein the *tet* operator-linked gene is a second transgene comprising a gene of interest operably linked to at least one *tet* operator sequence.

33. The organism of claim 32, wherein the at least one *tet* operator sequence is operatively linked upstream of the second transgene.

34. The organism of claim 32, wherein the at least one *tet* operator sequence is operatively linked downstream of the second transgene.

35. The organism of claim 21, wherein the *tet* operator-linked gene is an endogenous gene that has been operatively linked to at least one *tet* operator sequence.

36. The organism of claim 21, which is selected from the group consisting of: a mouse, a cow, a sheep, a pig, or a plant.--

REMARKS

Claim 1 was originally filed in the application and has now been canceled. New claims 21-36 have been added. Accordingly, claims 21-36 are pending.

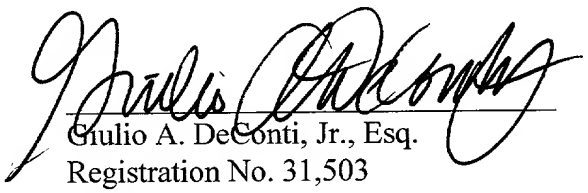
The title has been amended to more accurately reflect the subject being claimed in the application. The specification also has been amended to correct a number of minor informalities. New claims 21-36 are directed to transgenic organisms having tetracycline-regulated transcriptional regulatory systems. Support for these claims can be found in the specification at, for example, pages 17-19, 34 and 61-63.

No new matter has been added by way of the amendments to the specification or the new claims. Applicants respectfully request that these amendments be entered.

SUMMARY

All pending claims are believed to be in condition for allowance. If a telephone conversation with Applicants' Agent would expedite the prosecution of the above-identified application, the examiner is urged to call Applicants' Attorney at (617) 227-7400.

Respectfully submitted,



Giulio A. DeConti, Jr., Esq.
Registration No. 31,503
Attorney for Applicants

LAHIVE & COCKFIELD, LLP
28 State Street
Boston, MA 02109
Tel. (617) 227-7400
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